

2010 Sigma Xi Symposium Abstracts

Gustavus Adolphus College

April 30, 2010

Session Ia: Oral Papers

4:15 pm-6:30 pm

Nobel 201

4:15 pm The Geology of Seven Mile Creek Park

Jeremy Bock

Advisor: Laura Triplett

A geologic map is a fundamental scientific tool that shows the distribution of geologic material. This study used data gathered at Seven Mile Creek Park to make a geologic map. This data examines the relationships between ravine cutting, terrace development, and base level in a tributary to the Minnesota River, known as Seven Mile Creek. Within the Park there is an extensive network of ravines and there appear to be several levels of terraces. Having a geologic map lays the foundation for a more developed understanding of the regional geology and provides a concise information base for others to conduct work within the region.

4:30 pm Quantification of Glutathione using Fluorescence Detection

Siu On Auyeung

Advisor: Brenda Kelly

Glutathione (GSH) plays an important role in eukaryotes and some prokaryotes in cell redox maintenance and detoxification. In addition, the overexpression of GSH has been implicated in chemotherapeutic resistance for some cancers. The objective of this study was to establish an effective method to extract GSH from a J774A.1 mouse macrophage cell line and to quantify GSH levels using a derivitization reaction and fluorescence spectroscopy, preliminarily studied by students in the CHE-380 laboratory. The optimal incubation time and GSH concentration range (0-20 μM) for a linear GSH standard curve was established for the OPA and 2-aminophenol derivitization reaction. Different cell extraction methods were tested (mechanical vs. chemical cell lysis techniques) to extract and measure GSH levels in the cell line, however, concentrations were too low to fit within the current GSH standard curve. Work will continue to optimize reaction conditions for cell extraction and GSH detection.

4:45 pm Inhibition of Anti-microbial Products by Bacterial Signalling Molecules n-Acyl Homoserine Lactone

Eric Fitts

Advisor: John Lammert

Gram-negative bacteria make use of quorum sensing signaling molecules to detect cell density and regulate gene expression. These molecules have also been the recent focus of intense scrutiny for their potential in pharmacology. This study looks at the immunomodulatory effects that n-acyl homoserine lactones have on murine macrophage in vitro. (results pending on current experiment).

5:00 pm Understanding *Cellulomonas flavigena* through Genome Annotation and Functional Genomics Studies

Stephanie Hardel

Advisor: Kimberly Murphy

Cellulomonas flavigena is a gram-positive bacterium capable of cellulose and polysaccharide metabolism. *C. flavigena* is of great interest for bioenergy research as a potential source of novel cellulases. However, its mechanism for polysaccharide metabolism is not fully understood, and little is known regarding its metabolic and functional capabilities. We have initiated research to better understand metabolism of polysaccharides in *C. flavigena*. This includes annotation of its genome and functional analyses of several genes of interest.

5:15-5:30 pm Break

5:30 pm Non-Contact Measurements of Operating Deflection Shapes Using a Roving Ultrasound Transducer

Bradley Abell

Advisor: Tom Huber

This research developed a non-contact technique for measuring operating deflection shapes (ODS) stimulated by the ultrasound radiation force excitation from an air-coupled ultrasound transducer and measured using a laser Doppler vibrometer. Unlike conventional ODS measurements, this technique used a stationary vibrometer and roving ultrasound transducer. To demonstrate this method, a 0.80cm x 2.94cm brass cantilever was used to verify and compare to conventional ODS measurement techniques. A technique for fixing the soft edge that occurs when the ultrasound focal point falls off the edge of the cantilever was also developed during this experiment. This non-contact approach using a roving transducer allowed the laser Doppler vibrometer to remain stationary, proving to be an alternative method to the conventional scanning vibrometer used in ODS measurements.

5:45 pm Selectivity and Binding of Buthionine Sulfoximine Diastereomers to gamma-Glutamylcysteinylase

Benjamin Johnson-Tesch

Advisor: Brenda Kelly

Buthionine sulfoximine (BSO) has long been known to inhibit kinetic activity of gamma-glutamylcysteinylase (gamma-GCL). However, little data has been collected concerning the binding strength of BSO diastereomers to gamma-GCL and the selectivity of these BSO diastereomers. Using fluorescence spectroscopy, we have obtained evidence that suggests that the diastereomer L-S-BSO binds with greater affinity to gamma-GCL than the L-R-BSO diastereomer. Enzymatic activity, monitored using UV/Visible spectroscopy, shows the tightly bound L-S-BSO reduces gamma-GCL maximal velocity nearly 80%. The L-R-BSO, which showed weak binding, inhibited kinetic activity by less than 20% at the same concentration. We hypothesize the gamma-GCL binding site for BSO

sterically promotes selectivity for the L-S-BSO diastereomer.

6:00 pm Post-transcriptional Regulation of MPII

Gamachu Melkamu, Nick Guttormson, Krishan Jethwa

Advisors: Brenda Kelly and Jeff Dahlseid

MPII is a small cadmium-binding protein that is involved in the cellular detoxification of cadmium. It was previously observed that MPII protein levels were higher in cadmium treated cells, while the RNA levels remained the same (Demuyne et al. 2004). In response to this finding, our goal became to test the potential for post-transcriptional regulation of MPII. In order to test how MPII is regulated, MPII has been cloned into a heterologous system, Baker's yeast. By measuring cell growth over time for yeast strains with and without the MPII gene in media of varying cadmium levels, it has been shown that the MPII gene conveys a slight resistance to 8 μM Cadmium, shown by a greater rate of growth when compared to a wild type yeast control. However, the most significant finding was that the wild type yeast strain with the inserted MPII gene showed significant resistivity to 32 μM cadmium.

6:15 pm Selective Excitation of Cantilevers Using Ultrasound

Bryce Bork

Advisor: Tom Huber

Most conventional excitation techniques require physical contact with a mechanical shaker. We discuss a non-contact technique using the ultrasound radiation force. An audio-range modulation signal is combined with a 440-kHz carrier and emitted from two ultrasound transducers. The audio frequency range encompassed nearly overlapping transverse and torsional vibrational modes of a cantilever. A phase difference was introduced between the two transducers and allowed for the resolution of the two modes, which is not possible using conventional mechanical excitation.

Session Ib: Poster Presentations (in association with the Celebration of Creative Inquiry)

5:00 pm-7:00 pm

Jackson Campus Center

Photochemical Degradation Products of Imazethapyr in Natural Waterways

Annie Opseth

Advisor: Amanda Nienow

Imazethapyr is an herbicide in the imidazolinone family used to kill weeds in bean crops. As soybeans are a major crop throughout southern Minnesota and imazethapyr is considered slightly toxic by the EPA, its

environmental fate in local waterways is of importance. Degradation products of imazamox, another imidazolinone, have been studied using LC-MS. Our goal is to identify imazethapyr's degradation products, and add to the knowledge of the fate of imidazolinones in aquatic systems.

School vs. Home Lunches and Diet Variance: Project Healthy Schools Study

Cydni Smith

Only 6% of the 30.6 million school lunches served in the US daily meet requirements (mean sodium twice recommended level and 80% contain >30% calories from fat). Little is known regarding habits and outcomes of school lunch consuming children. 1297 middle-school students completed dietary recall including consumption of fruits and vegetables, fatty foods, and sugary drinks. Physiological measures included height, weight and a lipid panel. Children consuming school lunches were more likely to be overweight/obese (38.3% vs. 24.4%, $p < 0.001$). Children who regularly ate school lunch also reported higher intake of fatty meats (25.8% vs. 11.4%, $p < 0.001$), and sugary drinks (36% vs. 14.5%, $p < 0.001$), while reporting lower intake of fruits (24.9% vs. 31.3%, $p < 0.001$) and vegetables (16.3% vs. 91.2%, $p = 0.03$). Low density lipid cholesterol was also higher (94.0 vs. 87.5, $p < 0.01$). These data suggest an association between regular consumption of school lunches, less healthy eating habits and higher LDL.

Design of patient-tailored therapies using multiple holistic treatments to reduce stress indicators

Ashley Baumann, Becky Dove, Jeanifer Poon

Advisor: Sanjive Qazi

The central goal of Holistic Medicine, developed from thousands of years of whole body understanding and practice in Eastern Medicine, is to activate the body's biochemical response to heal and protect itself under periods of physiological stress. We propose to study the effects of holistic treatments, including Yoga and dance on college students stress levels by measuring specific stress indicators for multi-modal therapies. A mechanistic understanding from the endocrinological response to these treatments will allow for specific, subject-tailored design; the future goal being to optimize treatments to attain the greatest reduction in a panel of indicators measured.

Dependence of myohemerythrin folding on iron oxidation state

Alysha Dicke

Advisor: Brandy Russell

To better understand how oxidation states of metal atoms affect metalloprotein folding, unfolding thermodynamics of myohemerythrin (myoHr) were characterized. Two unfolding transitions for metmyoHr (Fe³⁺) were observed. The data suggest the first transition involves metmyoHr unfolding, and the second transition involves disruption to the diiron site. Furthermore, comparison of these data to the published unfolding data of deoxymyoHr (Fe²⁺) (Zhang, et al (1992) PNAS 89:7068) indicates that metmyoHr is more stable.

Histone 3.3A Affects Progeny Numbers and Sperm Storage in *Drosophila melanogaster*

Stephanie Snyder

Advisor: Margaret Bloch Qazi

Reproductive physiology is a critical component of an organism's reproductive success. Female sperm storage, a

commonly overlooked stage in reproduction, occurs when sperm are retained in specific regions of the female's reproductive tract. Identifying and characterizing genes involved in female sperm storage can better help us understand its role in the reproductive process. A genetic screen of the pomace fly, *Drosophila melanogaster*, identified Histone 3.3A as having a possible role in female reproduction. We characterized this gene by examining its role in components of female reproduction including: the number of eggs laid, progeny produced, and sperm storage as well as reproductive tract morphology (seminal receptacle length and spermathecae volume). Histone 3.3A influences female progeny production and sperm storage. Therefore mutant Histone 3.3A affects female *D. melanogaster* sperm storage which can lead to changes in reproduction.

Development of Multidimensional Chromatography for Trace Analysis in Complex Mixtures: Analysis of Phenytoin in Wastewater Treatment Plant Effluent and Hydrocortisone in Human Urine

Scott Simpkins, Michael Swenson, Steve Groskreutz

Advisor: Dwight Stoll

The occurrence of pharmaceuticals in the environment and the identification of certain biomarker molecules in biological fluids are issues of increasing analytical interest. Traditional analytical approaches to the measurement of these compounds in complex matrices are expensive and deficient in some aspects of analytical performance. Here we describe the development of a highly selective three dimensional liquid chromatographic method to analyze phenytoin and hydrocortisone without extensive sample preparation. We find that the method is versatile, cost effective, and quantitatively accurate, compared to existing methods.

Development of Rapid Liquid Chromatography-Based Methods for the Quantitation of Opiates and Benzodiazepines in Human Biofluids

Jonna Berry and Tomas Liskutin

Advisor: Dwight Stoll

The primary objective of this work is to develop rapid high performance liquid chromatography (HPLC) methods for the analyses of abused drugs in the benzodiazepine and opiate classes. Existing methods involve long analysis times (55 minutes per sample), and extensive sample preparation. Preliminary experimental results obtained using recently commercialized materials for HPLC are the most promising thus far. We have also developed a modeling strategy that will provide an efficient route to optimization of HPLC parameters after a complete set of experimental data has been collected.

Intratissue Refractive Index Shaping (IRIS) affects the Structure and Organization of Collagen Fibrils in the Corneal Stroma-An Electron Microscopy Study

Melissa Mackley

Advisor: Krystel R. Huxlin, University of Rochester, NY

Huxlin et al. developed a method for altering the refractive index (RI) of living, transparent corneal tissue without tissue destruction, a process termed intra-tissue refractive index shaping (IRIS). This mechanism was investigated through a quantitative assessment of IRIS effects on the size, spacing, and striation patterns of corneal collagen fibrils. Fibril area, center-to-center spacing, and dark band striation length was measured. IRIS

significantly decreased center-to-center spacing between collagen fibrils compared to control tissue. Fibril area was also significantly increased at the IRIS lines compared to fibrils outside the line sets. However, IRIS did not significantly alter collagen fibril striation length. These results could explain a localized densification of the collagen in the corneal tissue, which locally increases corneal RI.

Allosteric Behavior of Monomeric gamma-Glutamylcysteine Ligase in the Presence of Non-substrate Analogs

Chelsea Koepsell

Advisor: Brenda Kelly

The enzyme gamma-glutamylcysteine ligase (gamma-GCL) catalyzes the first and rate-limiting step in the synthesis of glutathione. Because of the wide reaching importance of glutathione and our limited structural knowledge of this enzyme, our objective was to determine the oligomeric state of E. coli gamma-GCL. Size exclusion chromatography and analysis through native gel electrophoresis indicate that E. coli gamma-GCL is monomeric. From these results, in conjunction with kinetics and fluorescence data which suggest that gamma-GCL exhibits positive cooperativity in the presence of non-substrate analogs, we hypothesize that there are two distinct binding sites within the monomeric enzyme. It is thought that these two sites can bind either a cysteine substrate or non-substrate analog, and that the presence of one filled site significantly impacts binding at the other site, leading to unusual enzyme kinetic patterns.